



Complete Set of Claims as Amended (37 C.F.R. § 1.121(c)(3))

1. A process for the isolation of nucleic acids from a sample including the following steps:
  - (a) applying at least one nucleic acid sample to a membrane;
  - (b) immobilizing the nucleic acids on the membrane;
  - (c) releasing the immobilized nucleic acids from the membrane; and
  - (d) removing the released nucleic acids through the membrane, whereby the membrane is comprised of one or more materials selected from the group consisting of nylon, polysulfone, polyethersulfone, polycarbonate, polyacrylate, acrylic copolymer, polyurethane, polyamide, polyvinylchloride, polyfluorocarbonate, poly-tetrafluoro-ethylene, polyvinylidene fluoride, polyethylene-tetrafluoro-ethylene-copolymerisate, polybenzimidazole, polyethylene-chlorotrifluoro-ethylene-copolymerisate, polyimide, polyphenylene sulfide, cellulose, cellulose-mix ester, cellulose nitrate, cellulose acetate, polyacrylnitrile, polyacrylnitril-copolymer, nitrocellulose, polypropylene and polyester.
2. The process according to Claim 1, characterized in that the nucleic acid sample is applied on top of the membrane and the nucleic acids are removed from below the membrane.
3. The process according to Claims 1 or 2, characterized in that the membrane is placed in a container with an inlet and outlet and the membrane fills the entire cross-section of the container, separating said inlet and outlet.
4. The process according to Claim 1, characterized in that the membrane is coated.
5. The process according to Claim 4, characterized in that the membrane has been made hydrophobic by the coating.
6. The process according to Claim 4, characterized in that the membrane has been made hydrophilic by the coating.

7. The process according to Claim 1, characterized in that the membrane is less than 1 mm thick.
8. The process according to Claim 7, characterized in that the membrane is less than 0.5 mm.
9. A process for the isolation of nucleic acids from a sample comprising the following steps:
  - (a) applying at least one nucleic acid sample to a surface;
  - (b) immobilizing the nucleic acids on the surface;
  - (c) releasing the immobilized nucleic acids from the surface with an elution agent, characterized in that the release takes place at a temperature  $T$ , whereby  $10^{\circ}\text{C} \geq T \geq T_{s,EM}$ , and  $T_{s,EM}$  equals the freezing point of the elution agent.
10. The process according to Claim 9, characterized in that the release takes place at temperature  $T$ , in which  $10^{\circ}\text{C} \geq T \geq 5^{\circ}\text{C}$ .
11. The process according to Claims 9, characterized in that the release takes place at temperature  $T$ , in which  $10^{\circ}\text{C} \geq T \geq 0^{\circ}\text{C}$ .
12. The process according to Claims 9, characterized in that the release takes place at temperature  $T$ , in which  $10^{\circ}\text{C} \geq T \geq -5^{\circ}\text{C}$ .
13. The process according to Claim 9, characterized in that the release takes place at temperature  $T$ , in which  $5^{\circ}\text{C} \geq T \geq T_{s,EM}$ .
14. A process for the isolation of nucleic acids from a sample comprising the following steps:
  - (a) adjusting a nucleic acid sample to binding conditions that permit immobilization of the nucleic acids contained in the sample on a surface;
  - (b) applying the nucleic acids sample to the surface; and
  - (c) immobilizing the nucleic acids on the surface, characterized in that, before and/or after adjusting the binding conditions there is a pre-

treatment of the sample.

15. The process according to Claim 14, characterized in that the pre-treatment takes place by salting out.
16. The process according to Claim 14, characterized in that the pre-treatment takes place by way of filtration, centrifugation, enzymatic treatment, temperature effect, precipitation, extraction, homogenization, mechanical reduction and/or binding of contaminants to surfaces.
17. The process according to Claim 14, characterized in that the binding conditions permit immobilization of RNA.
18. The process according to Claim 14, characterized in that the binding conditions permit immobilization of DNA.
19. The process according to Claim 14, characterized in that the following additional steps are included:
  - releasing of immobilized nucleic acids from the surface, and
  - removing the released nucleic acids from the surface.
20. The process according to any one of Claims 1, 9 or 14, characterized in that after the release step at least one additional step takes place:
  - performing at least one chemical reaction with the nucleic acids.
37. The process according to any one of Claims 9 or 14, characterized in that the sample is introduced onto the top of the surface.
38. A process according to any one of Claims 1, 9, or 19, characterized in that the immobilized nucleic acids are subjected to a washing step which takes place with at least one washing buffer after the immobilization and before any release steps.

39. The process according to Claim 38, characterized in that the washing step consists of the following steps for each washing buffer:
- applying a predetermined quantity of washing buffer on the surface; and
  - passing the washing buffer through the surface.
40. The process according to any one of Claims 1, 9, or 19, characterized in that an aqueous salt or buffer solution is used to release the nucleic acids.
41. The process according to any one of Claims 1, 9, or 19, characterized in that water is used to release the nucleic acids.
42. The process according to one of Claims 1, 9, or 14, characterized in that the application and immobilization of the nucleic acids includes the following steps:  
mixing at least one nucleic acid-containing sample with an immobilization buffer;  
applying said at least one nucleic acid-containing sample with the immobilization buffer to the surface or membrane; and  
passing the liquid components through the surface in essentially the same direction they were added.
43. The process according to any one of Claims 1, 9, or 19, characterized in that at least one of the steps is carried out by an automatic device, in a fully automatic manner.
44. The process according to Claim 43, characterized in that all steps of the process are performed by an automatic apparatus in a controlled sequence.
45. The process according to Claim 43, characterized in that a majority of nucleic acid isolations or reactions take place simultaneously.

46. The process according to any one of Claims 1, 9, or 19, characterized in that aqueous salt solutions of metal and/or ammonium cations with mineral acids are used to immobilize the nucleic acids.

47. The process according to Claim 46, wherein the aqueous salt solutions are of alkaline halides, alkaline-earth halides, alkaline sulfates, alkaline-earth sulfates, alkaline phosphates, alkaline-earth phosphates, or mixtures thereof.

48. The process according to Claim 47, characterized in that sodium halides, lithium halides and/or potassium halides and/or magnesium sulfate are used to immobilize the nucleic acids.

49. The process according to any one of Claims 1, 9, or 19, characterized in that aqueous solutions of salts of mono or polybasic or polyfunctional organic acids with alkaline or alkaline-earth metals are used to immobilize the nucleic acids.

50. The process according to Claim 49, characterized in that aqueous solutions of sodium, potassium or magnesium salts with organic dicarboxylic acids are used to immobilize the nucleic acids.

51. The process according to Claim 50, characterized in that the organic dicarboxylic acid is oxalic acid, malonic acid and/or succinic acid.

52. The process according to Claim 49, characterized in that aqueous solutions of sodium or potassium salts with a hydroxy or polyhydroxycarboxylic acid are used to immobilize the nucleic acids.

53. The process according to Claim 52, characterized in that the polyhydroxycarboxylic acid is citric acid.

54. The process according to any one of Claims 1, 9, or 19, characterized in that hydroxy-functional compounds of aliphatic or acyclic saturated or unsaturated hydrocarbons are used for the immobilization of the nucleic acids.

55. The process according to Claim 54, wherein said hydroxy-functional compounds are selected from the C<sub>1</sub>-C<sub>5</sub> alkanols.

56. The process according to Claim 55, wherein said alkanols are selected from methanol, ethanol, n-propanol, tert.-butanol, pentanols, and mixtures thereof.

57. The process according to Claim 54, wherein said hydroxy-functional compound is an alditol.

58. The process according to any one of Claims 1, 9, or 19, characterized in that a phenol or polyphenol is used for the immobilization of the nucleic acids.

59. The process according to any one of Claims 1, 9, or 19, wherein at least one chaotropic agent is used for the immobilization of the nucleic acids.

60. The process according to Claim 59, characterized in that the chaotropic agent is a salt selected from the group of trichloroacetates, thiocyanates, perchlorates, iodides, guanidinium hydrochloride, guanidinium isothiocyanate, and urea.

61. The process according to Claim 59, characterized in that 0.01 molar to 10 molar aqueous solutions of at least one chaotropic agent by itself, or in combination with other salts, is used to immobilize the nucleic acids.

62. The process according to Claim 61, characterized in that 0.1 molar to 7 molar aqueous solutions of at least one chaotropic agent by itself, or in combination with other salts, is used to immobilize the nucleic acids.

63. The process according to Claim 62, characterized in that 0.2 molar to 5 molar aqueous solutions of at least one chaotropic agent by itself, or in combination with other salts, is used to immobilize the nucleic acids.
64. The process according to Claim 61, wherein the chaotropic agent is selected from an aqueous solution of one or more of sodium perchlorate, guanidinium hydrochloride, guanidinium isothiocyanate, sodium iodide and potassium iodide.
65. The process according to Claim 38, wherein washing steps are carried out using salt or buffer solutions selected from aqueous salt solutions of metal and/or ammonium cations with mineral acids, including alkaline halides, alkaline-earth halides, alkaline sulfates, alkaline-earth sulfates, alkaline phosphates, alkaline-earth phosphates, or mixtures thereof; aqueous solutions of salts of mono or polybasic or polyfunctional organic acids with alkaline or alkaline-earth metals, including sodium, potassium or magnesium salts of organic dicarboxylic acids including oxalic acid, malonic acid and succinic acid; aqueous solutions of sodium or potassium salts of a hydroxy or polyhydroxycarboxylic acid including citric acid; hydroxy-functional compounds of aliphatic or acyclic saturated or unsaturated hydrocarbons including C<sub>1</sub>-C<sub>5</sub> alkanols and alditols; phenols or polyphenols; one or more chaotropic agents including salts selected from the group of trichloroacetates, thiocyanates, perchlorates, iodides, guanidinium hydrochloride, guanidinium isothiocyanate, and urea.
66. The process according to any one of Claims 9, 14, or 19, characterized in that the surface is a membrane.
67. The process according to Claim 66, characterized in that the membrane is a hydrophobic membrane.
68. The process according to Claim 67, characterized in that the hydrophobic membrane consists of a polymer with polar groups.

69. The process according to Claim 67, characterized in that the membrane is a hydrophilic membrane with a hydrophobic surface.

70. The process according to Claim 67, characterized in that the membrane is made of nylon, a polysulfone, polyethersulfone, polycarbonate, polypropylene, polyacrylate, acrylic copolymer, polyurethane, polyamide, polyvinylchloride, polyfluorocarbonate, poly-tetrafluoro-ethylene, polyvinylidene fluoride, polyethylene-tetrafluoro-ethylene-copolymerisate, a polyethylene-chlorotrifluoro-ethylene-copolymerisate, cellulose acetate, nitrocellulose, polybenzimidazole, polyimide, polyacrylonitrile, polyacrylonitrile-copolymer, cellulose-mix ester, cellulose nitrate, or polyphenylene sulfide.

71. The process according to Claim 70, characterized in that the membrane consists of hydrophobic nylon.

72. The process according to Claim 71, characterized in that the membrane is coated with a hydrophobizing coating agent selected from the group of paraffins, waxes, metal soaps, optionally containing additives selected from the group of aluminum or zirconium salts, quaternary organic compounds, ureic derivatives, lipid modified resins, silicones, zinc organic compounds and glutaric dialdehyde.

73. The process according to Claim 1, wherein the membrane is a hydrophilic membrane or a membrane made hydrophilic by pre-treatment.

74. The process according to Claim 73 characterized in that the membrane consists of hydrophilized nylon, polyethersulfone, polycarbonate, polyacrylate, acrylic copolymer, polyurethane, polyamide, polyvinylchloride, polyfluorocarbonate, poly-tetrafluoro-ethylene, polyvinylidene fluoride, polyethylene-tetrafluoro-ethylene-copolymerisate, a polyethylene-chlorotrifluoro-ethylene-copolymerisate, cellulose acetate, polypropylene, nitrocellulose, polybenzimidazole, polyimide, polyacrylonitrile, polyacrylonitrile-copolymer, cellulose-mix ester, polyester, polysulfone, cellulose nitrate, or polyphenylene sulfide.



75. The process according to any one of Claims 9, 14, or 19, characterized in that the membrane has a pore diameter of 0.001 to 50 micrometer.
76. The process according to any one of Claims 9, 14, or 19, characterized in that the surface is a hydrophobic fleece.
77. A process for isolating nucleic acids including the following steps:
- (a) providing an isolation device with at least one membrane located therein;
  - (b) applying a nucleic acid-containing sample to the isolation device;
  - (c) precipitating the nucleic acids contained in the sample with an alcohol, so that the nucleic acids are bound to the at least one membrane,
- characterized in that the pore size of said at least one membrane is equal or larger than 0.2 micrometer.
78. The process according to Claim 77, characterized in that the alcohol is added to the nucleic acid-containing sample prior to adding the sample to the isolation device.
79. The process according to Claim 77, characterized in that the alcohol is added to the nucleic acid-containing sample after adding the sample to the isolation device.
80. The process according to Claim 77, characterized in that the surface of the membrane is selected so that all the nucleic acids contained in the solution can be bound to the membrane.
81. The process according to Claim 77, wherein said membrane has a pore size equal to or greater than 0.2 micrometer.
82. The process according to Claim 81, wherein said nucleic acids precipitated are DNA and/or RNA.
83. The process according to Claim 77, wherein the alcohol used is a C<sub>1</sub>-C<sub>5</sub> alkanol with.

84. The process according to Claim 77, wherein the alcohol is isopropanol, and the volume ratio of the nucleic acids-containing sample to isopropanol is 2:1 to 1:1.
85. The process according to Claim 77, wherein the membrane is a hydrophobic membrane.
86. The process according to Claim 85, wherein the hydrophobic membrane consists of a polymer with polar groups.
87. The process according to Claim 85, wherein the membrane is a hydrophilic membrane with a hydrophobic surface.
88. The process according to Claim 85, characterized in that the membrane consists of nylon, polyethersulfone, polypropylene, polycarbonate, polyacrylate, acrylic copolymer, polyurethane, polyamide, polyvinylchloride, polyfluorocarbonate, poly-tetrafluoro-ethylene, polyvinylidene fluoride, polyethylene-tetrafluoro-ethylene-copolymerisate, a polyethylene-chlorotrifluoro-ethylene-copolymerisate or, polyphenylene sulfide.
89. The process according to Claim 88, characterized in that the membrane consists of a hydrophobic nylon.
90. The process according to Claim 87, characterized in that the membrane is coated with a waterproofing agent selected from the group of paraffins, waxes, metal soaps, optionally containing additives selected from aluminum or zirconium salts, quaternary organic compounds, ureic derivatives, lipid modified melamine resins, silicones, zinc organic compounds and/or glutaric dialdehyde.
91. The process according to Claim 77, wherein the membrane is a hydrophilic membrane or a hydrophilized membrane.
92. The process according to Claim 91, wherein the membrane consists of hydrophilized nylon, polyethersulfone, polycarbonate, polyacrylate, acrylic copolymer, polyurethane,

polyamide, polyvinylchloride, polyfluorocarbonate, poly-tetrafluoro-ethylene, polyvinylidene fluoride, polyethylene-tetrafluoro-ethylene-copolymerisate, a polyethylene-chlorotrifluoroethylene-copolymerisate, cellulose acetate, cellulose nitrate, or polyphenylene sulfide.

93. The process according to Claim 92, wherein the membrane consists of cellulose acetate or cellulose nitrate.

94. The process according to Claim 91, wherein the membrane has a pore size of more than 0.45  $\mu\text{m}$ .

95. The process according to Claim 91, wherein the membrane has a pore size of more than 0.6  $\mu\text{m}$ .

112. A method for isolating nucleic acids comprising contacting a sample containing nucleic acids with a material selected from the group of cellulose acetate; non-carboxylized, hydrophobic polyvinylidene fluoride; and massive, hydrophobic polytetrafluoroethylene.

113. The method of Claim 112, wherein said material is used in the form of a membrane.

114. The method of Claim 112, wherein said material is used in the form of a granulate.

115. The method of Claim 112, wherein the material is used in the form of a fiber.

116. The method of Claim 115, wherein the fibers are organized as a fleece.

121. The process according to Claim 62, wherein the chaotropic agent is selected from an aqueous solution of one or more of sodium perchlorate, guanidinium hydrochloride, guanidinium isothiocyanate, sodium iodide and potassium iodide.

122. The process according to Claim 63, wherein the chaotropic agent is selected from an aqueous solution of one or more of sodium perchlorate, guanidinium hydrochloride, guanidinium isothiocyanate, sodium iodide and potassium iodide.
123. The process according to Claim 68, characterized in that the membrane is a hydrophilic membrane with a hydrophobic surface.
124. The process according to Claim 68, characterized in that the membrane is made of nylon, a polysulfone, polyethersulfone, polycarbonate, polypropylene, polyacrylate, acrylic copolymer, polyurethane, polyamide, polyvinylchloride, polyfluorocarbonate, poly-tetrafluoro-ethylene, polyvinylidene fluoride, polyethylene-tetrafluoro-ethylene-copolymerisate, a polyethylene-chlorotrifluoro-ethylene-copolymerisate, cellulose acetate, nitrocellulose, polybenzimidazole, polyimide, polyacrylnitrile, polyacrylnitrile-copolymer, cellulose-mix ester, cellulose nitrate, or polyphenylene sulfide.